

1642



P#15 1600

## RAW SEQUENCE LISTING

DATE: 10/31/2002

PATENT APPLICATION: US/09/619,310B

TIME: 16:05:24

Input Set : A:\3759-0106P.ST25.txt

Output Set: N:\CRF4\10312002\I619310B.raw

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ENTERED

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3 <110> APPLICANT: THASTRUP, Ole
4   TULLIN, Soren
5   POULSEN, Lars K
6   BJORN, Sara P
8 <120> TITLE OF INVENTION: Novel Florescent Proteins
10 <130> FILE REFERENCE: 3759-0106P
12 <140> CURRENT APPLICATION NUMBER: 09/619,310B
13 <141> CURRENT FILING DATE: 2000-07-19
15 <150> PRIOR APPLICATION NUMBER: US 08/819,612
16 <151> PRIOR FILING DATE: 1997-03-17
18 <150> PRIOR APPLICATION NUMBER: PCT/DK96/00051
19 <151> PRIOR FILING DATE: 1996-01-31
21 <150> PRIOR APPLICATION NUMBER: DK 1065/95
22 <151> PRIOR FILING DATE: 1995-09-22
24 <160> NUMBER OF SEQ ID NOS: 23
26 <170> SOFTWARE: PatentIn version 3.1
28 <210> SEQ ID NO: 1
29 <211> LENGTH: 36
30 <212> TYPE: DNA
31 <213> ORGANISM: Artificial Sequence
33 <220> FEATURE:
34 <223> OTHER INFORMATION: GFP2 primer directed to A. victoria
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42 <212> TYPE: DNA
43 <213> ORGANISM: Artificial Sequence
45 <220> FEATURE:
46 <223> OTHER INFORMATION: GFP-1 primer directed to A. victoria
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49 aagaattcgg atcccttttag tgtcaattgg aagtct      36
52 <210> SEQ ID NO: 3
53 <211> LENGTH: 67
54 <212> TYPE: DNA
55 <213> ORGANISM: Artificial Sequence
57 <220> FEATURE:
58 <223> OTHER INFORMATION: 5' PCR primer incorporating the Y66H substitution responsible
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59   changing green fluorecence into blue fluorecence
61 <400> SEQUENCE: 3
62 ctacctgttc catggccaac gcttgctact actttcctca tgggtgttcaa tgcttttcta      60
64 gataccc                                          67
67 <210> SEQ ID NO: 4

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68 <211> LENGTH: 36  
69 <212> TYPE: DNA  
70 <213> ORGANISM: Artificial Sequence  
72 <220> FEATURE:  
73 <223> OTHER INFORMATION: 3' PCR primer incorporating the Y66H substitution responsible  
for  
74 changing green fluorescence into blue fluorescence  
76 <400> SEQUENCE: 4  
77 aagaattcgg atcccttag tgtcaattgg aagtct 36  
80 <210> SEQ ID NO: 5  
81 <211> LENGTH: 30  
82 <212> TYPE: DNA  
83 <213> ORGANISM: Artificial Sequence  
85 <220> FEATURE:  
86 <223> OTHER INFORMATION: 5' primer used to flank the Y66H-GFP  
88 <400> SEQUENCE: 5  
89 aattggtacc aaggaggtaa gctttatgag 30  
92 <210> SEQ ID NO: 6  
93 <211> LENGTH: 30  
94 <212> TYPE: DNA  
95 <213> ORGANISM: Artificial Sequence  
97 <220> FEATURE:  
98 <223> OTHER INFORMATION: 3' primer used to flank the Y66H-GFP  
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101 ctttcgtttt gaattcggat cccttagtg 30  
104 <210> SEQ ID NO: 7  
105 <211> LENGTH: 48  
106 <212> TYPE: DNA  
107 <213> ORGANISM: Artificial Sequence  
109 <220> FEATURE:  
110 <223> OTHER INFORMATION: large NcoI-XbaI vector fragment and ligated to SEQ ID NO:8  
112 <400> SEQUENCE: 7  
113 catggccaac gctgtgcaact actctctctt atggtgttca atgctttt 48  
116 <210> SEQ ID NO: 8  
117 <211> LENGTH: 48  
118 <212> TYPE: DNA  
119 <213> ORGANISM: Artificial Sequence  
121 <220> FEATURE:  
122 <223> OTHER INFORMATION: synthetic NcoI-XbaI DNA linker and ligated to SEQ ID NO:7  
124 <400> SEQUENCE: 8  
125 ctagaaaagc attgaacacc ataagagaga gtagtgacaa gcgttggc 48  
128 <210> SEQ ID NO: 9  
129 <211> LENGTH: 48  
130 <212> TYPE: DNA  
131 <213> ORGANISM: Artificial Sequence  
133 <220> FEATURE:  
134 <223> OTHER INFORMATION: large NcoI-XbaI vector fragment ligated to SEQ ID NO:10  
136 <400> SEQUENCE: 9  
137 catggccaac gctgtgcaact actctcactt atggtgttca atgctttt 48  
140 <210> SEQ ID NO: 10

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141 <211> LENGTH: 48
142 <212> TYPE: DNA
143 <213> ORGANISM: Artificial Sequence
145 <220> FEATURE:
146 <223> OTHER INFORMATION: NcoI-XbaI DNA linker ligated to SEQ ID NO:9
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153 <211> LENGTH: 36
154 <212> TYPE: DNA
155 <213> ORGANISM: Artificial Sequence
157 <220> FEATURE:
158 <223> OTHER INFORMATION: 5' primer based on pGFP-N1 plasmid
160 <400> SEQUENCE: 11
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164 <210> SEQ ID NO: 12
165 <211> LENGTH: 35
166 <212> TYPE: DNA
167 <213> ORGANISM: Artificial Sequence
169 <220> FEATURE:
170 <223> OTHER INFORMATION: 3' primer based on pGFP-N1 plasmid
172 <400> SEQUENCE: 12
173 gaatcgtaga tctttatttg tatagttcat ccatg      35
176 <210> SEQ ID NO: 13
177 <211> LENGTH: 40
178 <212> TYPE: DNA
179 <213> ORGANISM: Artificial Sequence
181 <220> FEATURE:
182 <223> OTHER INFORMATION: 5' primer based on pZeoSV-LacZ plasmid
184 <400> SEQUENCE: 13
185 tggaataagc tttatggatc ccgctgtttt acaacgtcgt      40
188 <210> SEQ ID NO: 14
189 <211> LENGTH: 34
190 <212> TYPE: DNA
191 <213> ORGANISM: Artificial Sequence
193 <220> FEATURE:
194 <223> OTHER INFORMATION: 3' primer based on pZeoSV-LacZ plasmid
196 <400> SEQUENCE: 14
197 gcgcgaattc ttattattat ttttgacacc agac      34
200 <210> SEQ ID NO: 15
201 <211> LENGTH: 764
202 <212> TYPE: DNA
203 <213> ORGANISM: Aequorea Victoria
205 <220> FEATURE:
206 <221> NAME/KEY: CDS
207 <222> LOCATION: (8)..(721)
208 <223> OTHER INFORMATION:
W--> 211 <400> 15
      212 aagcttt atg agt aaa gga gaa gaa ctt ttc act gga gtt gtc cca att      49

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213      Met Ser Lys Gly Glu Glu Leu Phe Thr Gly Val Val Pro Ile
214      1      5      10
216 ctt gtt gaa tta gat ggc gat gtt aat ggg caa aaa ttc tct gtt agt      97
217 Leu Val Glu Leu Asp Gly Asp Val Asn Gly Gln Lys Phe Ser Val Ser
218 15      20      25      30
220 gga gag ggt gaa ggt gat gca aca tac gga aaa ctt acc ctt aaa ttt      145
221 Gly Glu Gly Glu Gly Asp Ala Thr Tyr Gly Lys Leu Thr Leu Lys Phe
222      35      40      45
224 att tgc act act ggg aag cta cct gtt cca tgg cca acg ctt gtc act      193
225 Ile Cys Thr Thr Gly Lys Leu Pro Val Pro Trp Pro Thr Leu Val Thr
226      50      55      60
228 act ttc tct tat ggt gtt caa tgc ttt tca aga tac cca gat cat atg      241
229 Thr Phe Ser Tyr Gly Val Gln Cys Phe Ser Arg Tyr Pro Asp His Met
230      65      70      75
232 aaa cag cat gac ttt ttc aag agt gcc atg ccc gaa ggt tat gta cag      289
233 Lys Gln His Asp Phe Phe Lys Ser Ala Met Pro Glu Gly Tyr Val Gln
234      80      85      90
236 gaa aga act ata ttt tac aaa gat gac ggg aac tac aag aca cgt gct      337
237 Glu Arg Thr Ile Phe Tyr Lys Asp Asp Gly Asn Tyr Lys Thr Arg Ala
238 95      100      105      110
240 gaa gtc aag ttt gaa ggt gat acc ctt gtt aat aga atc gag tta aaa      385
241 Glu Val Lys Phe Glu Gly Asp Thr Leu Val Asn Arg Ile Glu Leu Lys
242      115      120      125
244 ggt att gat ttt aaa gaa gat gga aac att ctt gga cac aaa atg gaa      433
245 Gly Ile Asp Phe Lys Glu Asp Gly Asn Ile Leu Gly His Lys Met Glu
246      130      135      140
248 tac aac tat aac tca cat aat gta tac atc atg gca gac aaa cca aag      481
249 Tyr Asn Tyr Asn Ser His Asn Val Tyr Ile Met Ala Asp Lys Pro Lys
250      145      150      155
252 aat gga atc aaa gtt aac ttc aaa att aga cac aac att aaa gat gga      529
253 Asn Gly Ile Lys Val Asn Phe Lys Ile Arg His Asn Ile Lys Asp Gly
254      160      165      170
256 agc gtt caa tta gca gac cat tat caa caa aat act cca att ggc gat      577
257 Ser Val Gln Leu Ala Asp His Tyr Gln Gln Asn Thr Pro Ile Gly Asp
258 175      180      185      190
260 ggc cct gtc ctt tta cca gac aac cat tac ctg tcc acg caa tct gcc      625
261 Gly Pro Val Leu Leu Pro Asp Asn His Tyr Leu Ser Thr Gln Ser Ala
262      195      200      205
264 ctt tcc aaa gat ccc aac gaa aag aga gat cac atg atc ctt ctt gag      673
265 Leu Ser Lys Asp Pro Asn Glu Lys Arg Asp His Met Ile Leu Leu Glu
266      210      215      220
268 ttt gta aca gct gct ggg att aca cat ggc atg gat gaa cta tac aaa      721
269 Phe Val Thr Ala Ala Gly Ile Thr His Gly Met Asp Glu Leu Tyr Lys
270      225      230      235
272 taaatgtcca gacttccaat tgacactaaa gggatccgaa ttc      764
275 <210> SEQ ID NO: 16
276 <211> LENGTH: 238
277 <212> TYPE: PRT
278 <213> ORGANISM: Aequorea Victoria

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280 <400> SEQUENCE: 16
282 Met Ser Lys Gly Glu Glu Leu Phe Thr Gly Val Val Pro Ile Leu Val
283 1 5 10 15
286 Glu Leu Asp Gly Asp Val Asn Gly Gln Lys Phe Ser Val Ser Gly Glu
287 20 25 30
290 Gly Glu Gly Asp Ala Thr Tyr Gly Lys Leu Thr Leu Lys Phe Ile Cys
291 35 40 45
294 Thr Thr Gly Lys Leu Pro Val Pro Trp Pro Thr Leu Val Thr Thr Phe
295 50 55 60
298 Ser Tyr Gly Val Gln Cys Phe Ser Arg Tyr Pro Asp His Met Lys Gln
299 65 70 75 80
302 His Asp Phe Phe Lys Ser Ala Met Pro Glu Gly Tyr Val Gln Glu Arg
303 85 90 95
306 Thr Ile Phe Tyr Lys Asp Asp Gly Asn Tyr Lys Thr Arg Ala Glu Val
307 100 105 110
310 Lys Phe Glu Gly Asp Thr Leu Val Asn Arg Ile Glu Leu Lys Gly Ile
311 115 120 125
314 Asp Phe Lys Glu Asp Gly Asn Ile Leu Gly His Lys Met Glu Tyr Asn
315 130 135 140
318 Tyr Asn Ser His Asn Val Tyr Ile Met Ala Asp Lys Pro Lys Asn Gly
319 145 150 155 160
322 Ile Lys Val Asn Phe Lys Ile Arg His Asn Ile Lys Asp Gly Ser Val
323 165 170 175
326 Gln Leu Ala Asp His Tyr Gln Gln Asn Thr Pro Ile Gly Asp Gly Pro
327 180 185 190
330 Val Leu Leu Pro Asp Asn His Tyr Leu Ser Thr Gln Ser Ala Leu Ser
331 195 200 205
334 Lys Asp Pro Asn Glu Lys Arg Asp His Met Ile Leu Leu Glu Phe Val
335 210 215 220
338 Thr Ala Ala Gly Ile Thr His Gly Met Asp Glu Leu Tyr Lys
339 225 230 235

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342 &lt;210&gt; SEQ ID NO: 17

343 &lt;211&gt; LENGTH: 764

344 &lt;212&gt; TYPE: DNA

345 &lt;213&gt; ORGANISM: Aequorea victoria

347 &lt;220&gt; FEATURE:

348 &lt;221&gt; NAME/KEY: CDS

349 &lt;222&gt; LOCATION: (8)..(724)

350 &lt;223&gt; OTHER INFORMATION:

W--&gt; 353 &lt;400&gt; 17

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354 aagcttt atg agt aaa gga gaa gaa ctt ttc act gga gtt gtc cca att 49
355 Met Ser Lys Gly Glu Glu Leu Phe Thr Gly Val Val Pro Ile
356 1 5 10
358 ctt gtt gaa tta gat ggc gat gtt aat ggg caa aaa ttc tcc gtt agt 97
359 Leu Val Glu Leu Asp Gly Asp Val Asn Gly Gln Lys Phe Ser Val Ser
360 15 20 25 30
362 gga gag ggt gaa ggt gat gca aca tac gga aaa ctt acc ctt aaa ttt 145
363 Gly Glu Gly Glu Gly Asp Ala Thr Tyr Gly Lys Leu Thr Leu Lys Phe
364 35 40 45

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## VERIFICATION SUMMARY

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TIME: 16:05:25

Input Set : A:\3759-0106P.ST25.txt

Output Set: N:\CRF4\10312002\I619310B.raw

L:211 M:258 W: Mandatory Feature missing, <223> Blank for SEQ#:15,Line#:208  
L:353 M:258 W: Mandatory Feature missing, <223> Blank for SEQ#:17,Line#:350  
L:495 M:258 W: Mandatory Feature missing, <223> Blank for SEQ#:19,Line#:492  
L:651 M:258 W: Mandatory Feature missing, <223> Blank for SEQ#:21,Line#:648